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PATENT

## IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of: MARTIN *et al.*

Serial No.: 10/026,302

Filed: December 19, 2001

For: Crown Ether Derivatives

Examiner: Bruck Kifle

Group Art Unit: 1624

Docket No.: MP.0070

MARKED-UP VERSION OF THE CLAIMSCommissioner for Patents  
U.S. Patent and Trademark Office  
PO Box 1450  
Alexandria, VA 22313-1450

Dear Sir:

This Marked-up Version of the Claims is being submitted along with the Response to the Office Action dated September 3, 2004. These Marked-up Claims are being submitted on or before the three (3) month extended due date of March 3, 2005. A Petition for Extension of Time is also enclosed (*See*, Transmittal).

The Examiner is respectfully requested to enter the following Claim amendments.

## CERTIFICATE OF TRANSMISSION

I HEREBY CERTIFY THAT THIS PAPER AND THE DOCUMENTS REFERRED AS BEING ATTACHED OR ENCLOSED HERewith ARE BEING  
FACSIMILE TRANSMITTED TO THE UNITED STATES PATENT AND TRADEMARK OFFICE ON 3/3/05 TO 1.703.872.9305  
By [Signature]

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**Claims:**

Claims 1-25. (Canceled).

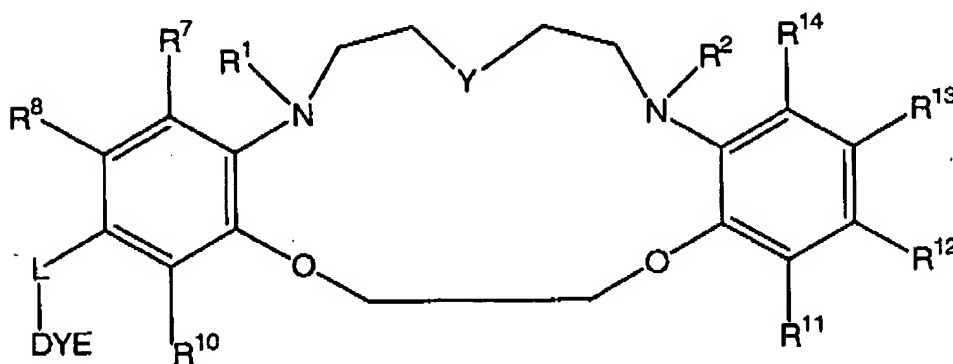
26. (Currently Amended) ~~A composition, as claimed in Claim 25~~ The composition according to Claim 80, wherein said compound is substituted by only one -L-R<sub>x</sub>, or -L-S<sub>c</sub>, that is bound at R<sup>8</sup>, R<sup>9</sup>, R<sup>12</sup>, or R<sup>13</sup>.
27. (Currently Amended) ~~A composition, as claimed in Claim 25~~ The composition according to Claim 80, wherein R<sup>1</sup> and R<sup>2</sup> are C<sub>1</sub>-C<sub>6</sub> alkyl that are substituted one or more times by cyano, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>.
28. (Currently Amended) ~~A composition, as claimed in Claim 27~~ The composition according to Claim 80, wherein R<sup>8</sup> and R<sup>9</sup>, and optionally R<sup>12</sup> and R<sup>13</sup>, taken in combination, form a fused DYE that is a substituted or unsubstituted benzofuran.
29. (Currently Amended) ~~A composition, as claimed in Claim 25~~ The composition according to Claim 80, wherein said compound is substituted by exactly two DYE or fused DYE moieties.
30. (Currently Amended) ~~A composition, as claimed in Claim 25~~ The composition according to Claim 80, wherein said compound is substituted by exactly one -L-DYE moiety at R<sup>9</sup>, and said compound is optionally substituted by exactly one -L-R<sub>x</sub> or exactly one -L-S<sub>c</sub> at a position other than R<sup>9</sup>.
31. (Currently Amended) ~~A composition, as claimed in Claim 24~~ The composition according to Claim 80, wherein each L of the compound is independently a single covalent bond, or a covalent linkage that is linear or branched, cyclic or heterocyclic, saturated or unsaturated, having 1-20 nonhydrogen atoms selected from the group consisting of C, N, P, O and S; and are composed of any combination of ether, thioether, amine, ester, carboxamide, sulfonamide, hydrazide bonds and aromatic or heteroaromatic bonds.
32. (Currently Amended) ~~A composition, as claimed in Claim 31~~ The composition according to Claim 80, wherein each L of the compound is a single covalent bond or has the

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formula  $-(CH_2)_d(CONH(CH_2)_e)_z-$  or  $-O(CH_2)_d(CONH(CH_2)_e)_z-$ , where  $d$  is an integer from 0-5,  $e$  is an integer from 1-5 and  $z$  is 0 or 1.

33. (Canceled).
34. (Currently Amended) ~~A composition, as claimed in Claim 26~~ The composition according to Claim 80, wherein said compound is substituted by exactly one  $S_{CT}$  ~~which  $S_C$  that~~ is a protein, a polysaccharide, a biotin, or a silica.
35. (Currently Amended) ~~A composition, as claimed in Claim 26~~ The composition according to Claim 80, wherein said compound is substituted by exactly one  $R_x$  selected from the group consisting of a succinimidyl ester of a carboxylic acid, a haloacetamide, a hydrazine, an isothiocyanate, a maleimide, an aliphatic amine, a silyl halide, and a psoralen.
36. (Currently Amended) ~~A composition, as claimed in Claim 24~~ The composition according to Claim 80, where the compound has the formula:



wherein  $Y$  is  $O$  or  $NR^4$ .

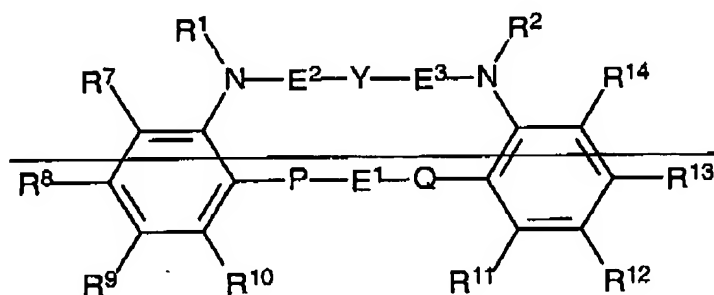
37. (Original) A composition, as claimed in Claim 36, wherein each DYE on the compound is a fluorescein, a rhodamine, a rhodol, a polyazaindacene, an oxazine, a 3H-xanthen-6-ol-3-one, a 6-amino-3H-xanthen-3-one, or a 6-amino-3H-xanthen-3-imine.

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38. (Previously Amended) A composition, as claimed in Claim 37, wherein  $R^1$  and  $R^2$  are  $C_1$ - $C_8$  alkyl that are substituted one or more times by  $-(C=O)-O-R^{16}$  or  $-(C=O)-NR^{17}R^{18}$ .
39. (Original) A composition, as claimed in Claim 38, wherein  $R^1$  and  $R^2$  are  $C_1$ - $C_8$  alkyl that are substituted one or more times by  $-(C=O)-O-R^{16}$ , where each  $R^{16}$  is H, an alpha-acyloxymethyl, a t-butyldimethyldimethylsilyl, or a biologically compatible salt.
40. (Currently Amended) A composition, as claimed in Claim 36, ~~further comprising a~~ wherein the metal ion that is  $Ca^{2+}$ ,  $Na^+$ ,  $K^+$ , or  $Zn^{2+}$  ~~associated with said compound.~~
41. (Canceled).
42. (Canceled).
43. (Canceled).
44. (Currently Amended) A method of detecting a target cationic metal ion in a sample, comprising:

a) adding to said sample, in an amount sufficient to generate a detectable optical response when said target ion is present, a compound having the formula:



wherein

~~P and Q are independently O, S, or  $NR^3$ , where each  $R^3$  is independently H or  $C_1$ - $C_8$  alkyl;~~

~~Y is O, S, or  $NR^4$ , where  $R^4$  is H; or is  $-L-R_x$ ,  $-L-S_G$ , or  $-L-DYE$ ; or is  $C_1$ - $C_8$  alkyl or an aryl or heteroaryl ring system, which alkyl or ring system is optionally substituted by~~

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~~halogen, azido, nitro, nitroso, amino, C<sub>1</sub>-C<sub>8</sub> alkylamino, C<sub>2</sub>-C<sub>12</sub> dialkylamino, cyano, L-R<sub>X</sub>, L-S<sub>C</sub>, or L-DYE; or by C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy that is itself optionally substituted by halogen, amino, hydroxy, (SO<sub>2</sub>)R<sup>16</sup>, (SO<sub>2</sub>)OR<sup>16</sup>, (C=O)R<sup>16</sup>, (C=O)OR<sup>16</sup>, or (C=O)NR<sup>17</sup>R<sup>18</sup>; wherein~~

~~R<sup>16</sup> is H or C<sub>1</sub>-C<sub>8</sub> alkyl; or L-R<sub>X</sub>, L-S<sub>C</sub>, or L-DYE;~~

~~R<sup>16</sup> is H, a C<sub>1</sub>-C<sub>8</sub> alkyl, a benzyl, alpha-acyloxyalkyl and t-butyl dimethylsilyl, a biologically compatible salt; or L-R<sub>X</sub>, L-S<sub>C</sub>, or L-DYE;~~

~~R<sup>17</sup> and R<sup>18</sup> are independently H, C<sub>1</sub>-C<sub>8</sub> alkyl, C<sub>1</sub>-C<sub>6</sub> carboxyalkyl, an alpha-acyloxyalkyl, a t-butyl dimethylsilyl, or a biologically compatible salt; or L-R<sub>X</sub>, L-S<sub>C</sub>, or L-DYE; or R<sup>17</sup> and R<sup>18</sup> taken in combination form a 5- or 6-membered aliphatic ring that optionally incorporates an oxygen atom;~~

~~each L is independently a covalent linkage;~~

~~each R<sub>X</sub> is independently a reactive group;~~

~~each S<sub>C</sub> is independently a conjugated substance;~~

~~DYE is a chemical moiety with an absorption maximum beyond 320 nm;~~

~~E<sup>1</sup>, E<sup>2</sup>, and E<sup>3</sup> are independently (CR<sup>6</sup>)<sub>n</sub>, where n = 2, 3, 4, and each R<sup>6</sup> is independently H or CH<sub>3</sub>, or two R<sup>6</sup> moieties on adjacent carbons of one or more of E<sup>1</sup>, E<sup>2</sup> or E<sup>3</sup>, when taken in combination, form a 5- or 6-membered aliphatic ring;~~

~~R<sup>1</sup> and R<sup>2</sup> are independently L-R<sub>X</sub>, L-S<sub>C</sub>, or L-DYE; or C<sub>1</sub>-C<sub>18</sub> alkyl or C<sub>2</sub>-C<sub>18</sub> arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by (SO<sub>2</sub>)R<sup>16</sup>, (SO<sub>2</sub>)OR<sup>16</sup>, (C=O)R<sup>16</sup>, (C=O)OR<sup>16</sup>, (C=O)NR<sup>17</sup>R<sup>18</sup>; or by C<sub>1</sub>-C<sub>8</sub> alkylamino, C<sub>2</sub>-C<sub>12</sub> dialkylamino; or by C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, (SO<sub>2</sub>)R<sup>16</sup>, (SO<sub>2</sub>)OR<sup>16</sup>, (C=O)R<sup>16</sup>, (C=O)OR<sup>16</sup>, (C=O)NR<sup>17</sup>R<sup>18</sup>;~~

~~R<sup>7</sup>-R<sup>14</sup> are independently H, halogen, azido, nitro, nitroso, amino, cyano, L-R<sub>X</sub>, L-S<sub>C</sub>, L-DYE; or C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen,~~

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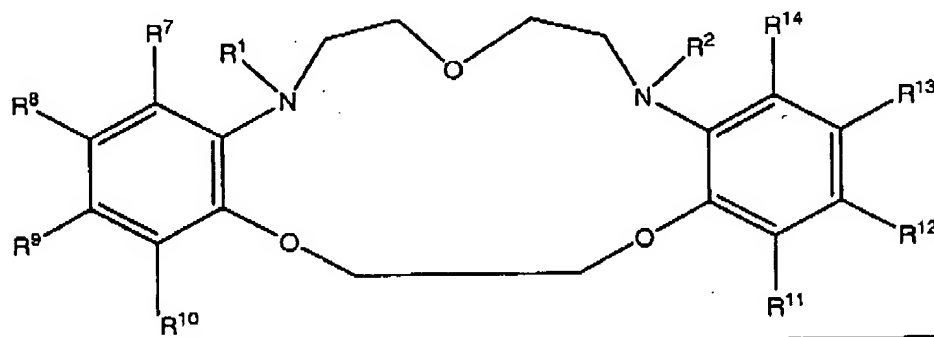
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amino, hydroxy,  $(\text{SO}_2)\text{-R}^{15}$ ,  $(\text{SO}_2)\text{-O-R}^{15}$ ,  $(\text{C=O})\text{-R}^{15}$ ,  $(\text{C=O})\text{-O-R}^{16}$ , or  $(\text{C=O})\text{-NR}^{17}\text{R}^{18}$ ;

or any two adjacent substituents  $\text{R}^7\text{-R}^{14}$ , taken in combination, form a fused six-membered benzo moiety, which is optionally substituted by halogen, azido, nitro, nitroso, amino, cyano,  $\text{-L-R}_x$ ,  $\text{-L-S}_c$ , or  $\text{-L-DYE}$ ; or  $\text{C}_1\text{-C}_6$  alkyl or  $\text{C}_1\text{-C}_6$  alkoxy, each of which is optionally substituted by halogen, amino, hydroxy,  $(\text{C=O})\text{-R}^{15}$ ,  $(\text{C=O})\text{-O-R}^{16}$ , or  $(\text{C=O})\text{-NR}^{17}\text{R}^{18}$ ;

or any two adjacent substituents  $\text{R}^7\text{-R}^{14}$ , taken in combination with each other, and with the aromatic ring they are bound to, form a fused DYE;

provided that said compound is substituted by at least one  $\text{-L-DYE}$  moiety at one or more of  $\text{R}^1$ ,  $\text{R}^2$ ,  $\text{R}^3$ , and  $\text{R}^7\text{-R}^{14}$ , or at least two of  $\text{R}^7\text{-R}^{14}$ , taken in combination, form a fused DYE;



wherein  $\text{R}^1$  is  $\text{-L-R}_x$ ,  $\text{-L-S}_c$ ,  $\text{-L-DYE}$ ;  $\text{C}_1\text{-C}_{18}$  alkyl or  $\text{C}_7\text{-C}_{18}$  arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by  $(\text{SO}_2)\text{-R}^{15}$ ,  $(\text{SO}_2)\text{-O-R}^{15}$ ,  $(\text{C=O})\text{-R}^{15}$ ,  $(\text{C=O})\text{-O-R}^{16}$ ,  $(\text{C=O})\text{-NR}^{17}\text{R}^{18}$ ; or by  $\text{C}_1\text{-C}_6$  alkylamino,  $\text{C}_2\text{-C}_{12}$  dialkylamino; or by  $\text{C}_1\text{-C}_6$  alkyl or  $\text{C}_1\text{-C}_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $(\text{SO}_2)\text{-R}^{15}$ ,  $(\text{SO}_2)\text{-O-R}^{15}$ ,  $(\text{C=O})\text{-R}^{15}$ ,  $(\text{C=O})\text{-O-R}^{16}$ ,  $(\text{C=O})\text{-NR}^{17}\text{R}^{18}$ ;

$\text{R}^2$  is  $\text{-L-R}_x$ ,  $\text{-L-S}_c$ ,  $\text{-L-DYE}$ ;  $\text{C}_1\text{-C}_{18}$  alkyl or  $\text{C}_7\text{-C}_{18}$  arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by  $(\text{SO}_2)\text{-R}^{15}$ ,  $(\text{SO}_2)\text{-O-R}^{15}$ ,  $(\text{C=O})\text{-R}^{15}$ ,  $(\text{C=O})\text{-O-R}^{16}$ ,  $(\text{C=O})\text{-NR}^{17}\text{R}^{18}$ ; or by  $\text{C}_1\text{-C}_6$  alkylamino,  $\text{C}_2\text{-C}_{12}$  dialkylamino; or by  $\text{C}_1\text{-C}_6$  alkyl or  $\text{C}_1\text{-C}_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $(\text{SO}_2)\text{-R}^{15}$ ,  $(\text{SO}_2)\text{-O-R}^{15}$ ,  $(\text{C=O})\text{-R}^{15}$ ,  $(\text{C=O})\text{-O-R}^{16}$ ,  $(\text{C=O})\text{-NR}^{17}\text{R}^{18}$ ;

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alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{16}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ ,  $-(C=O)-NR^{17}R^{18}$ .

wherein  $R^{16}$  is H,  $C_1-C_6$  alkyl,  $-L-R_x$ ,  $-L-S_c$ , or  $-L-DYE$ ;

$R^{16}$  is H, a  $C_1-C_6$  alkyl, a benzyl, alpha-acyloxyalkyl, t-butyldimethylsilyl, a biologically compatible salt,  $-L-R_x$ ,  $-L-S_c$ , or  $-L-DYE$ ;

$R^{17}$  is H,  $C_1-C_6$  alkyl,  $C_1-C_6$  carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt,  $-L-R_x$ ,  $-L-S_c$ , or  $-L-DYE$ ;

$R^{18}$  is H,  $C_1-C_6$  alkyl,  $C_1-C_6$  carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt,  $-L-R_x$ ,  $-L-S_c$ , or  $-L-DYE$ ;

or  $R^{17}$  and  $R^{18}$  taken in combination form a 5- or 6-membered aliphatic ring that optionally incorporates an oxygen atom;

L is a covalent linkage;

$R_x$  is an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a silyl halide, a sulfonyl halide, or a thiol;

$S_c$  is an amino acid, a peptide, a protein, a polysaccharide, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a synthetic polymer, a polymeric microparticle, a biological cell, a biotin, a silica or a virus;

DYE is a chemical moiety with an absorption maximum beyond 320 nm;

$R^7$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_x$ ,  $-L-S_c$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ .

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R<sup>8</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>8</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>9</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>8</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>10</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>8</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>11</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>8</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>12</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>8</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>13</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>8</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>14</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>8</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

or any two adjacent substituents R<sup>7</sup>-R<sup>14</sup>, taken in combination, form a fused six-membered benzo moiety, which is optionally substituted by halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>8</sub> alkoxy, each of which is optionally substituted by halogen, amino, hydroxy, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

or any two adjacent substituents R<sup>7</sup>-R<sup>14</sup>, taken in combination with each other, and with the aromatic ring they are bound to, form a fused DYE;



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provided that the compound is substituted by at least one -L-DYE, -L-R<sub>x</sub>, or -L-S<sub>c</sub> at R<sup>1</sup>, R<sup>2</sup>, R<sup>4</sup>, R<sup>7</sup>, R<sup>8</sup>, R<sup>9</sup>, R<sup>10</sup>, R<sup>11</sup>, R<sup>12</sup>, R<sup>13</sup>, or R<sup>14</sup>; or at least two of R<sup>7</sup>-R<sup>14</sup>, taken in combination, form a fused DYE;

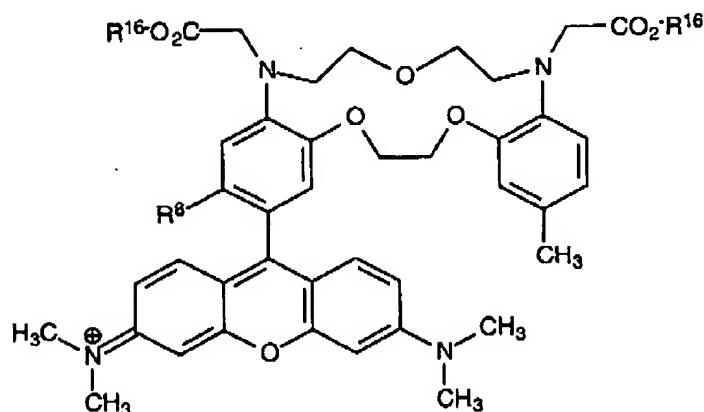
b) illuminating said sample to generate said detectable optical response whereby said target ion is present.

45. (Original) A method, as claimed in Claim 44, wherein said detectable optical response is a fluorescence response.

46. (Previously Amended) A method, as claimed in Claim 45, wherein said illuminating is performed in conjunction with a fluorometer, fluorescence microscope, laser scanner, flow cytometer, a microfluidic device, or a fiber optic probe.

47. (Original) A method, as claimed in Claim 44, wherein said target metal ion is Na<sup>+</sup>, K<sup>+</sup>, Ca<sup>2+</sup>, or Zn<sup>2+</sup>.

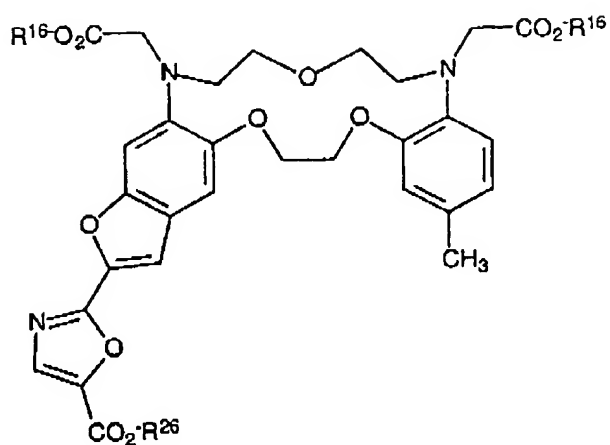
48. (Previously Amended) A method, as claimed in Claim 44, wherein said compound has the formula:



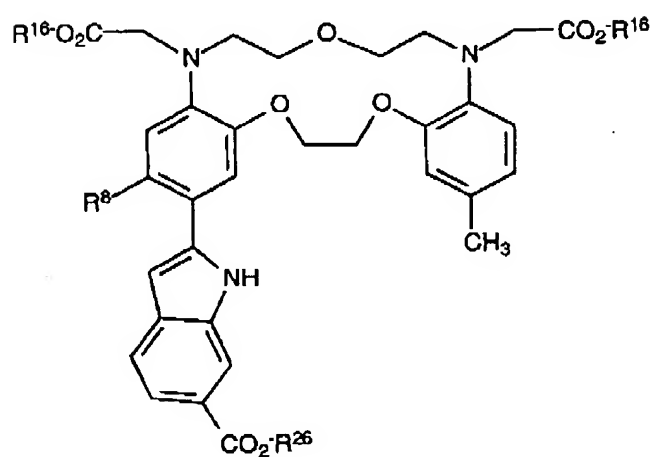
or the formula:

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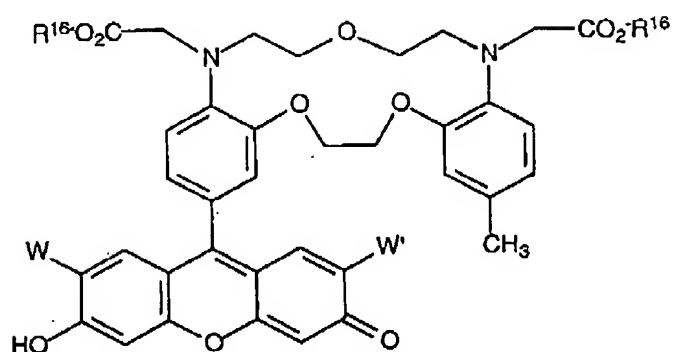
or the formula:



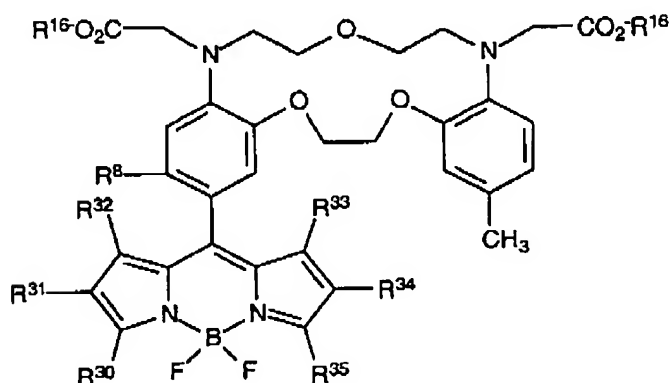
or the formula:

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or the formula:



wherein

$R^8$ , where present, is independently H or a  $C_1$ - $C_6$  alkoxy, which is optionally substituted by -  
(C=O)-O- $R^{16}$  or -(C=O)-NR $^{17}$ R $^{18}$ ;

$R^{16}$  and  $R^{26}$ , where present, are independently H, a  $C_1$ - $C_8$  alkyl, a benzyl, an alpha-acyloxyalkyl,  
a t-butyl(dimethylsilyl), or a biologically compatible salt;

$R^{17}$  and  $R^{18}$ , where present, are independently H, a  $C_1$ - $C_8$  alkyl,  $C_1$ - $C_8$  carboxyalkyl, or a  
biologically compatible salt;

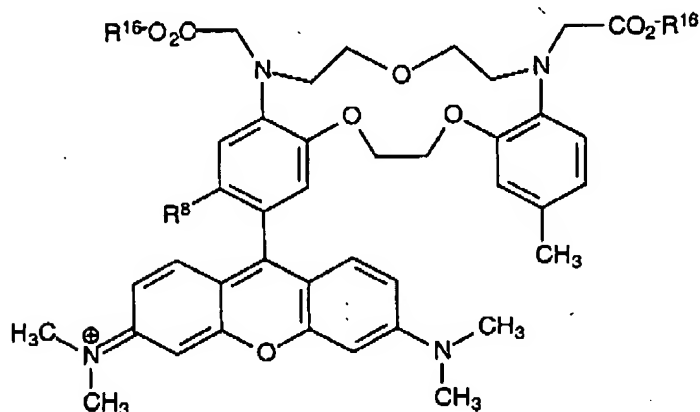
W and W', where present, are independently F or Cl;

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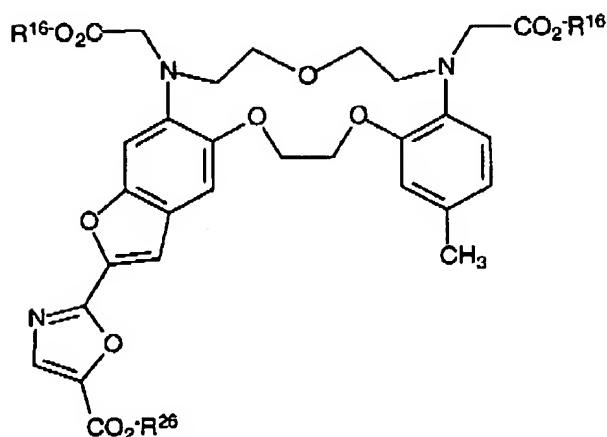
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$R^{30}$ - $R^{35}$ , where present, are independently H, halogen, nitro, sulfo, cyano, alkyl, perfluoroalkyl, alkoxy, alkenyl, alkynyl, cycloalkyl, arylalkyl, or acyl, wherein the alkyl portions of each contain fewer than 20 carbons; or an aryl or heteroaryl ring system.

49. (Original) A method, as claimed in Claim 48, wherein said target metal ion is  $Na^+$  or  $K^+$ .
50. (Previously Amended) A method, as claimed in Claim 44, wherein said sample comprises living cells or biological fluids.
51. (Previously Amended) A kit for the detection or quantification of a target metal ion, comprising a compound having the formula:



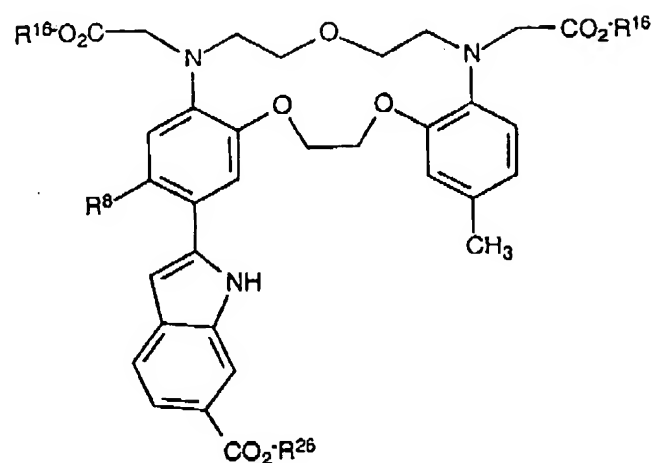
or the formula:



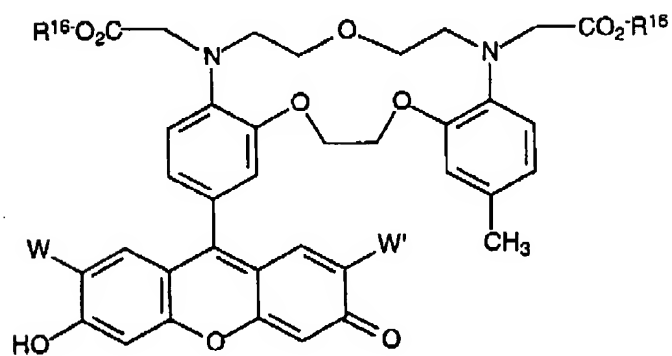
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or the formula:



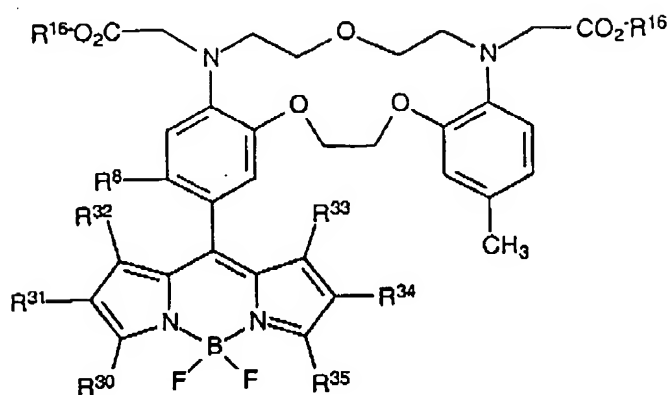
or the formula:



or the formula:

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wherein

$R^8$ , where present, is independently H or a  $C_1$ - $C_6$  alkoxy, which is optionally substituted by -  
(C=O)-O- $R^{18}$  or -(C=O)-NR $^{17}$  $R^{18}$ ;

$R^{16}$  and  $R^{26}$ , where present, are independently H, a  $C_1$ - $C_6$  alkyl, a benzyl, an alpha-acyloxyalkyl,  
a t-butyldimethylsilyl, or a biologically compatible salt;

$R^{17}$  and  $R^{18}$ , where present, are independently H, a  $C_1$ - $C_6$  alkyl,  $C_1$ - $C_6$  carboxyalkyl, or a  
biologically compatible salt;

W and W', where present, are independently F or Cl;

$R^{30}$ - $R^{35}$ , where present, are independently H, halogen, nitro, sulfo, cyano, alkyl, perfluoroalkyl,  
alkoxy, alkenyl, alkynyl, cycloalkyl, arylalkyl, or acyl, wherein the alkyl portions of each  
contain fewer than 20 carbons; or an aryl or heteroaryl ring system;

and comprising one or more components selected from the group consisting of:

- a) a calibration standard of a target ion;
- b) an ionophore;
- c) a fluorescence standard;
- d) an aqueous buffer solution; and

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e) an organic solvent.

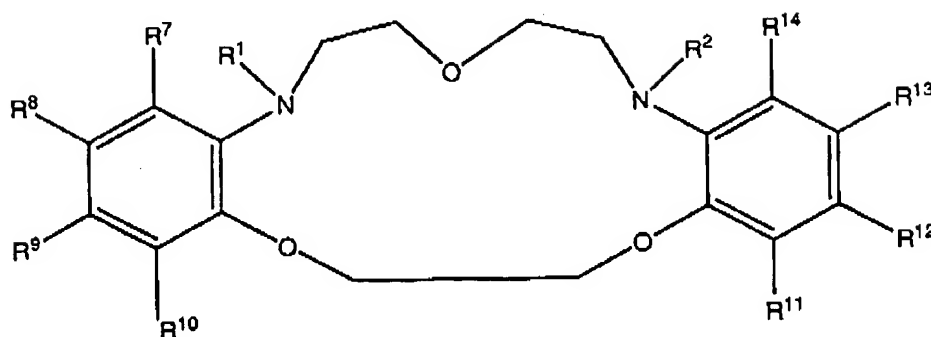
52. (Canceled).

53. (Canceled).

54. (Canceled).

55. (Canceled).

56. (New) A compound having the formula:



wherein  $R^1$  is  $-L-R_x$ ,  $-L-S_c$ ,  $-L-DYE$ ;  $C_1-C_{18}$  alkyl or  $C_7-C_{18}$  arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ ,  $-(C=O)-NR^{17}R^{18}$ ; or by  $C_1-C_6$  alkylamino,  $C_2-C_{12}$  dialkylamino; or by  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ ,  $-(C=O)-NR^{17}R^{18}$ ;

$R^2$  is  $-L-R_x$ ,  $-L-S_c$ ,  $-L-DYE$ ;  $C_1-C_{18}$  alkyl or  $C_7-C_{18}$  arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ ,  $-(C=O)-NR^{17}R^{18}$ ; or by  $C_1-C_6$  alkylamino,  $C_2-C_{12}$  dialkylamino; or by  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ ,  $-(C=O)-NR^{17}R^{18}$ ;

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wherein  $R^{15}$  is H,  $C_1$ - $C_6$  alkyl, -L- $R_x$ , -L- $S_c$ , or -L-DYE;

$R^{16}$  is H, a  $C_1$ - $C_6$  alkyl, a benzyl, alpha-acyloxyalkyl, t-butyldimethylsilyl, a biologically compatible salt, -L- $R_x$ , -L- $S_c$ , or -L-DYE;

$R^{17}$  is H,  $C_1$ - $C_8$  alkyl,  $C_1$ - $C_6$  carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt, -L- $R_x$ , -L- $S_c$ , or -L-DYE;

$R^{18}$  is H,  $C_1$ - $C_6$  alkyl,  $C_1$ - $C_6$  carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt, -L- $R_x$ , -L- $S_c$ , or -L-DYE;

or  $R^{17}$  and  $R^{18}$  taken in combination form a 5- or 6-membered aliphatic ring that optionally incorporates an oxygen atom;

L is a covalent linkage;

$R_x$  is an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a silyl halide, a sulfonyl halide, or a thiol;

$S_c$  is an amino acid, a peptide, a protein, a polysaccharide, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a synthetic polymer, a polymeric microparticle, a biological cell, a biotin, a silica or a virus;

DYE is a chemical moiety with an absorption maximum beyond 320 nm;

$R^7$  is H, halogen, azido, nitro, nitroso, amino, cyano, -L- $R_x$ , -L- $S_c$ , -L-DYE,  $C_1$ - $C_8$  alkyl or  $C_1$ - $C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{15}$ , or  $-(C=O)-NR^{17}R^{18}$ ;



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$R^8$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_X$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

$R^9$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_X$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

$R^{10}$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_X$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

$R^{11}$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_X$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

$R^{12}$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_X$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

$R^{13}$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_X$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

$R^{14}$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_X$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

or any two adjacent substituents  $R^7-R^{14}$ , taken in combination, form a fused six-membered benzo moiety, which is optionally substituted by halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_X$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is optionally substituted by halogen, amino, hydroxy,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

or any two adjacent substituents  $R^7-R^{14}$ , taken in combination with each other, and with the aromatic ring they are bound to, form a fused DYE;

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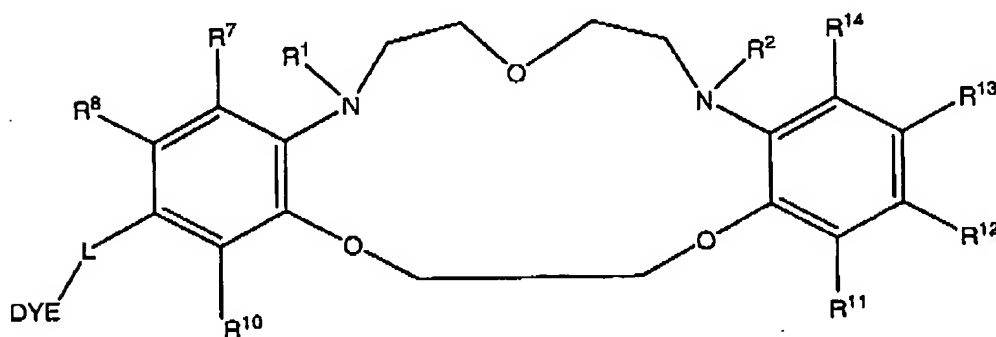
provided that the compound is substituted by at least one -L-DYE, -L-R<sub>x</sub>, or -L-S<sub>c</sub> at R<sup>1</sup>, R<sup>2</sup>, R<sup>4</sup>, R<sup>7</sup>, R<sup>8</sup>, R<sup>9</sup>, R<sup>10</sup>, R<sup>11</sup>, R<sup>12</sup>, R<sup>13</sup>, or R<sup>14</sup>; or at least two of R<sup>7</sup>-R<sup>14</sup>, taken in combination, form a fused DYE.

57. (New) The compound according to Claim 56, wherein the compound is substituted by only one -L-R<sub>x</sub>, or -L-S<sub>c</sub>, that is bound at R<sup>8</sup>, R<sup>9</sup>, R<sup>12</sup>, or R<sup>13</sup>.
58. (New) The compound according to Claim 56, wherein R<sup>1</sup> and R<sup>2</sup> are C<sub>1</sub>-C<sub>6</sub> alkyl that are substituted one or more times by cyano, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>.
59. (New) The compound according to Claim 58, wherein R<sup>16</sup>, R<sup>17</sup> and R<sup>18</sup> are independently H or C<sub>1</sub>-C<sub>6</sub> alkyl.
60. (New) The compound according to Claim 56, wherein R<sup>8</sup> and R<sup>9</sup>, and optionally R<sup>12</sup> and R<sup>13</sup>, taken in combination, form a fused DYE that is a substituted or unsubstituted benzofuran.
61. (New) The compound according to Claim 56, wherein the compound is substituted by exactly two DYE or fused DYE moieties.
62. (New) The compound according to Claim 56, wherein the compound is substituted by exactly one -L-DYE moiety at R<sup>9</sup>, and said compound is optionally substituted at a position other than R<sup>9</sup> by exactly one -L-R<sub>x</sub> or exactly one -L-S<sub>c</sub>.
63. (New) The compound according to Claim 56, wherein L is independently a single covalent bond, or a covalent linkage that is linear or branched, cyclic or heterocyclic, saturated or unsaturated, having 1-20 nonhydrogen atoms selected from the group consisting of C, N, P, O and S; and are composed of any combination of ether, thioether, amine, ester, carboxamide, sulfonamide, hydrazide bonds and aromatic or heteroaromatic bonds.
64. (New) The compound according to Claim 56, wherein L is a single covalent bond or has the formula -(CH<sub>2</sub>)<sub>d</sub>(CONH(CH<sub>2</sub>)<sub>e</sub>)<sub>z</sub>- or -O(CH<sub>2</sub>)<sub>d</sub>(CONH(CH<sub>2</sub>)<sub>e</sub>)<sub>z</sub>-, where d is an integer from 0-5, e is an integer from 1-5 and z is 0 or 1.

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65. (New) The compound according to Claim 56, wherein the DYE moiety is selected from the group consisting of indole, a coumarin, a stilbene, a xanthene, an oxazine, and a polyazaindacene.
66. (New) The compound according to Claim 56, wherein the compound is substituted by exactly one  $S_C$  that is a protein, a polysaccharide, a biotin, a synthetic polymer or a silica.
67. (New) The compound according to Claim 56, wherein the compound is substituted by at least one  $R_x$  selected from the group consisting of a succinimidyl ester of a carboxylic acid, a haloacetamide, a hydrazine, an isothiocyanate, a maleimide, an aliphatic amine, a silyl halide, and a psoralen.
68. (New) A compound having the formula:



wherein  $R^1$  is  $-L-R_x$ ,  $-L-S_C$ ,  $-L-DYE$ ;  $C_1-C_{18}$  alkyl or  $C_7-C_{18}$  arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{16}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ ,  $-(C=O)-NR^{17}R^{18}$ ; or by  $C_1-C_6$  alkylamino,  $C_2-C_{12}$  dialkylamino; or by  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{16}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ ,  $-(C=O)-NR^{17}R^{18}$ ;

$R^2$  is  $-L-R_x$ ,  $-L-S_C$ ,  $-L-DYE$ ;  $C_1-C_{18}$  alkyl or  $C_7-C_{18}$  arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{16}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ ,  $-(C=O)-NR^{17}R^{18}$ ;

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(C=O)-NR<sup>17</sup>R<sup>18</sup>; or by C<sub>1</sub>-C<sub>6</sub> alkylamino, C<sub>2</sub>-C<sub>12</sub> dialkylamino; or by C<sub>1</sub>-C<sub>8</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

wherein R<sup>15</sup> is H, C<sub>1</sub>-C<sub>6</sub> alkyl, -L-R<sub>x</sub>, -L-S<sub>c</sub>, or -L-DYE;

R<sup>16</sup> is H, a C<sub>1</sub>-C<sub>6</sub> alkyl, a benzyl, alpha-acyloxyalkyl, t-butyldimethylsilyl, a biologically compatible salt, -L-R<sub>x</sub>, -L-S<sub>c</sub>, or -L-DYE;

R<sup>17</sup> is H, C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>1</sub>-C<sub>6</sub> carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt, -L-R<sub>x</sub>, -L-S<sub>c</sub>, or -L-DYE;

R<sup>18</sup> is H, C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>1</sub>-C<sub>6</sub> carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt, -L-R<sub>x</sub>, -L-S<sub>c</sub>, or -L-DYE;

or R<sup>17</sup> and R<sup>18</sup> taken in combination form a 5- or 6-membered aliphatic ring that optionally incorporates an oxygen atom;

L is a covalent linkage;

R<sub>x</sub> is selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a silyl halide, a sulfonyl halide, or a thiol;

S<sub>c</sub> is selected from the group consisting of an amino acid, a peptide, a protein, a polysaccharide, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a drug, a hormone, a lipid, a lipid assembly, a synthetic polymer, a polymeric microparticle, a biological cell, a biotin, a silica and a virus;

DYE is selected from the group consisting of indole, a coumarin, a stilbene, a xanthene, an oxazine, a polyazaindacene, a benzofuran, a pyrene, an anthracene, a naphthalene, an acridine, a benzindole, an oxazole, a benzoxazole, a thiazole, a

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benzothiazole, a 4-amino-7-nitrobenz-2-oxa-1,3-diazole (NBD), a cyanine, a carbocyanine, a carbostyryl, a porphyrin, a salicylate, an anthranilate, an azulene, a perylene, a pyridine, a quinoline, a benzoxazine, a carbazine a phenalenone and a benzphenalenone;

R<sup>7</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>8</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>10</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>11</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>12</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>13</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>; and

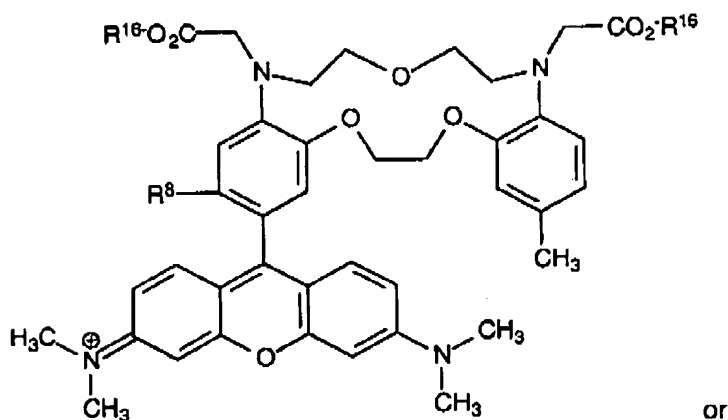
R<sup>14</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>X</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>.

69. (New) The compound according to Claim 68, wherein DYE is an indole, a coumarin, a stilbene, a xanthene, an oxazine, or a polyazaindacene.

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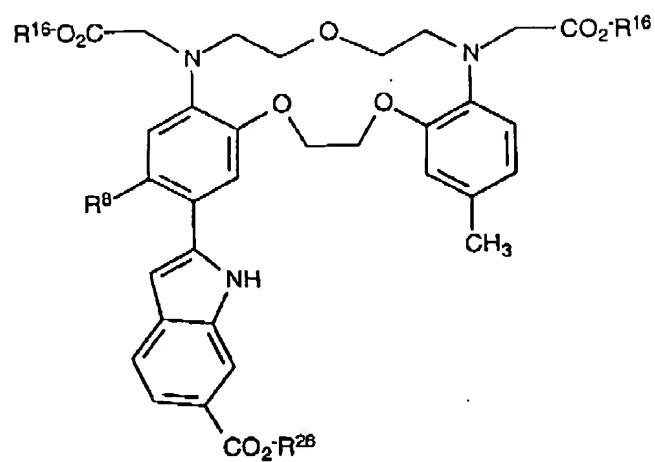
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70. (New) The compound according to Claim 68, wherein the xanthene is selected from the group consisting of a fluorescein, a rhodamine, a rhodol, a 3*H*-xanthen-6-ol-3-one, a 6-amino-3*H*-xanthen-3-one, and a 6-amino-3*H*-xanthen-3-imine; wherein L is a single covalent bond.
71. (New) The compound according to Claim 68, wherein R<sup>1</sup> and R<sup>2</sup> are C<sub>1</sub>-C<sub>6</sub> alkyl that are substituted one or more times by -(C=O)-O-R<sup>16</sup> or -(C=O)-NR<sup>17</sup>R<sup>18</sup>.
72. (New) The compound according to Claim 68, wherein R<sup>1</sup> and R<sup>2</sup> are C<sub>1</sub>-C<sub>6</sub> alkyl that are substituted one or more times by -(C=O)-O-R<sup>16</sup>, where each R<sup>16</sup> is H, C<sub>1</sub>-C<sub>8</sub> alkyl, an alpha-acyloxymethyl, a t-butyltrimethylsilyl, or a biologically compatible salt.
73. (New) The compound according to Claim 68, wherein the compound is

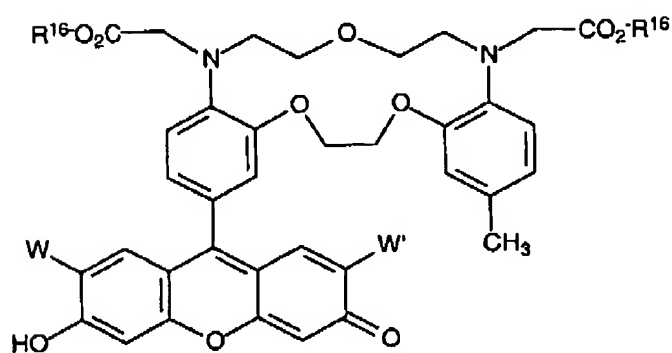


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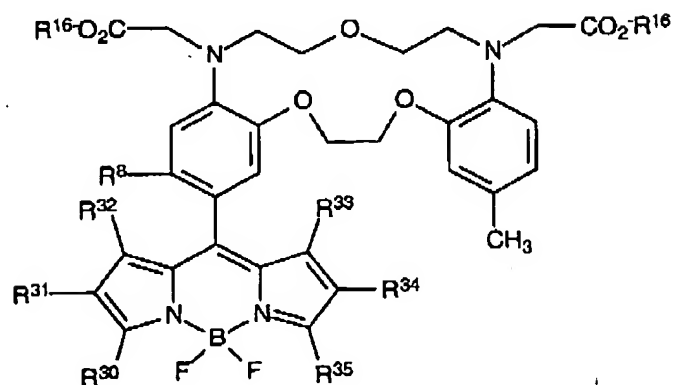
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or



or



wherein

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$R^8$ , where present, is independently H or a  $C_1$ - $C_6$  alkoxy, which is optionally substituted by -  
(C=O)-O- $R^{16}$  or -(C=O)-NR $^{17}$ R $^{18}$ ;

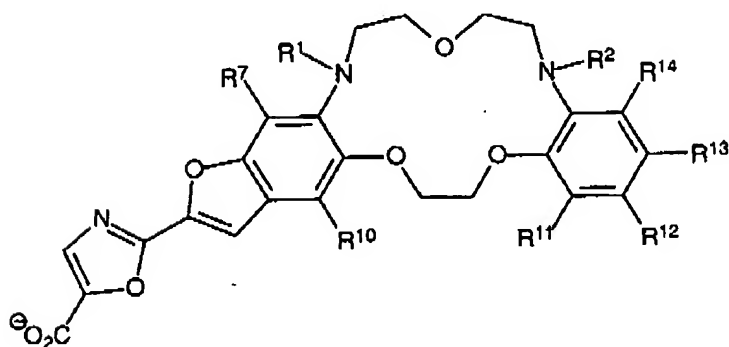
$R^{16}$  and  $R^{26}$ , where present, are independently H, a  $C_1$ - $C_6$  alkyl, a benzyl, an alpha-acyloxyalkyl,  
a t-butyldimethylsilyl, or a biologically compatible salt;

$R^{17}$  and  $R^{18}$ , where present, are independently H, a  $C_1$ - $C_6$  alkyl,  $C_1$ - $C_6$  carboxyalkyl, or a  
biologically compatible salt;

W and W', where present, are independently F or Cl;

$R^{30}$ - $R^{35}$ , where present, are independently H, halogen, nitro, sulfo, cyano, alkyl, perfluoroalkyl,  
alkoxy, alkenyl, alkynyl, cycloalkyl, arylalkyl, or acyl, wherein the alkyl portions of each  
contain fewer than 20 carbons; or an aryl or heteroaryl ring system; or adjacent  
substituents  $R^{31}$  and  $R^{32}$ , and adjacent substituents  $R^{33}$  and  $R^{34}$ , when taken in  
combination form a fused benzo ring that is optionally substituted by hydrogen, halogen,  
nitro, sulfo, cyano, alkyl, perfluoroalkyl, alkoxy, alkenyl, alkynyl, cycloalkyl, alkylthio,  
alkylamido, amino, monoalkylamino or dialkylamino wherein the alkyl portions of each  
contain fewer than 20 carbons.

74. (New) A compound having the formula



wherein  $R^1$  is -L- $R_X$ , -L- $S_C$ , -L-DYE;  $C_1$ - $C_{18}$  alkyl or  $C_7$ - $C_{18}$  arylalkyl, each of which is optionally  
substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or  
heteroaryl ring system; or by -(SO $_2$ )- $R^{15}$ , -(SO $_2$ )-O- $R^{15}$ , -(C=O)- $R^{15}$ , -(C=O)-O- $R^{16}$ , -



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(C=O)-NR<sup>17</sup>R<sup>18</sup>; or by C<sub>1</sub>-C<sub>6</sub> alkylamino, C<sub>2</sub>-C<sub>12</sub> dialkylamino; or by C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>2</sup> is -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE; C<sub>1</sub>-C<sub>18</sub> alkyl or C<sub>7</sub>-C<sub>18</sub> arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>16</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, -(C=O)-NR<sup>17</sup>R<sup>18</sup>; or by C<sub>1</sub>-C<sub>6</sub> alkylamino, C<sub>2</sub>-C<sub>12</sub> dialkylamino; or by C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

wherein R<sup>15</sup> is H, C<sub>1</sub>-C<sub>6</sub> alkyl, -L-R<sub>x</sub>, -L-S<sub>C</sub>, or -L-DYE;

R<sup>16</sup> is H, a C<sub>1</sub>-C<sub>6</sub> alkyl, a benzyl, alpha-acyloxyalkyl, t-butyldimethylsilyl, a biologically compatible salt, -L-R<sub>x</sub>, -L-S<sub>C</sub>, or -L-DYE;

R<sup>17</sup> is H, C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>1</sub>-C<sub>6</sub> carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt, -L-R<sub>x</sub>, -L-S<sub>C</sub>, or -L-DYE;

R<sup>18</sup> is H, C<sub>1</sub>-C<sub>6</sub> alkyl, C<sub>1</sub>-C<sub>6</sub> carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt, -L-R<sub>x</sub>, -L-S<sub>C</sub>, or -L-DYE;

or R<sup>17</sup> and R<sup>18</sup> taken in combination form a 5- or 6-membered aliphatic ring that optionally incorporates an oxygen atom;

L is a covalent linkage;

R<sub>x</sub> is selected from the group consisting of an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a silyl halide, a sulfonyl halide, or a thiol;

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Sc is selected from the group consisting of an amino acid, a peptide, a protein, a polysaccharide, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a drug, a hormone, a lipid, a lipid assembly, a synthetic polymer, a polymeric microparticle, a biological cell, a biotin, a silica and a virus;

DYE is selected from the group consisting of indole, a coumarin, a stilbene, a xanthene, an oxazine, a polyazaindacene, a benzofuran, a pyrene, an anthracene, a naphthalene, an acridine, a benzindole, an oxazole, a benzoxazole, a thiazole, a benzothiazole, a 4-amino-7-nitrobenz-2-oxa-1,3-diazole (NBD), a cyanine, a carbocyanine, a carbostyryl, a porphyrin, a salicylate, an anthranilate, an azulene, a perylene, a pyridine, a quinoline, a benzoxazine, a carbazine a phenalenone and a benzphenalenone;

R<sup>7</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>10</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>11</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>12</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>13</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>; and

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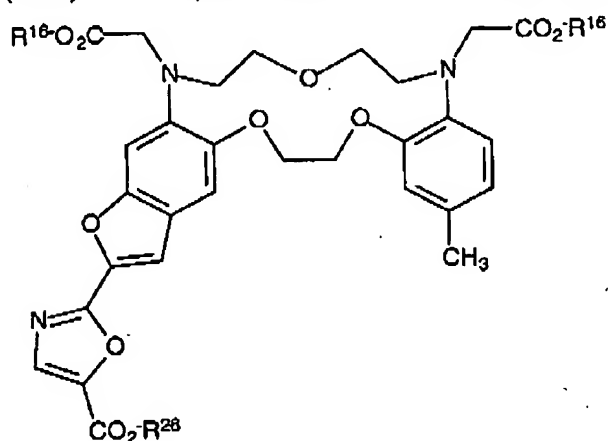
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$R^{14}$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_x$ ,  $-L-S_C$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ .

75. (New) The compound according to Claim 74, wherein  $R^1$  and  $R^2$  are  $C_1-C_6$  alkyl that are substituted one or more times by  $-(C=O)-O-R^{16}$  or  $-(C=O)-NR^{17}R^{18}$ .

76. (New) The compound according to Claim 74, wherein  $R^1$  and  $R^2$  are  $C_1-C_6$  alkyl that are substituted one or more times by  $-(C=O)-O-R^{16}$ , where each  $R^{16}$  is H,  $C_1-C_6$  alkyl, an alpha-acyloxymethyl, a t-butyldimethyldimethylsilyl, or a biologically compatible salt.

77. (New) The compound according to Claim 74, wherein the compound is

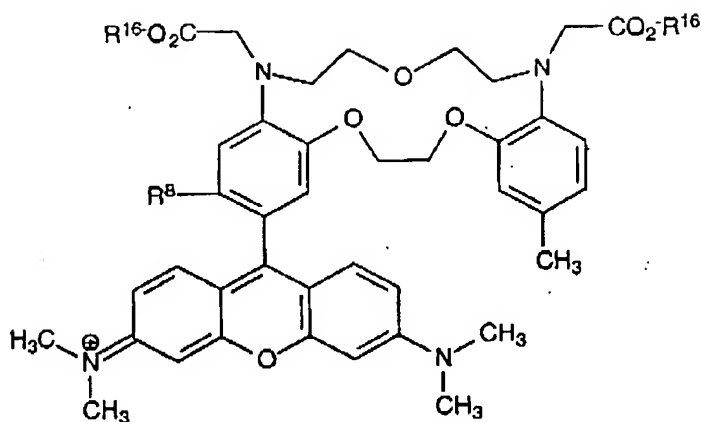


wherein  $R^{16}$  and  $R^{26}$  are independently H, a  $C_1-C_6$  alkyl, a benzyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, or a biologically compatible salt.

78. A compound having the formula:

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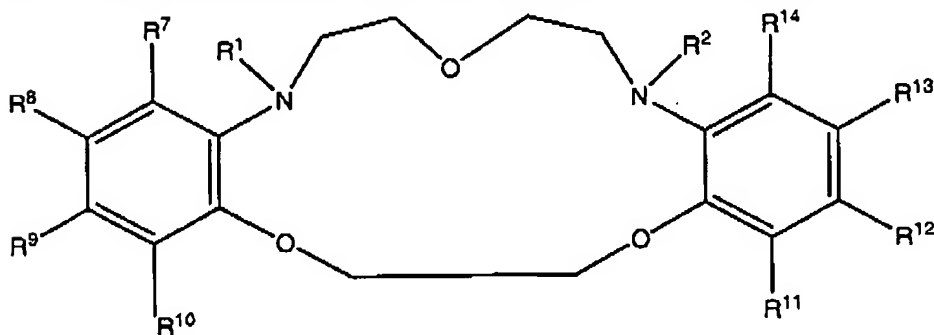
and its salts, wherein

$R^8$ , is H or a  $C_1$ - $C_6$  alkoxy, which is optionally substituted by  $-(C=O)-O-R^{16}$  or  $-(C=O)-NR^{17}R^{18}$ ;

$R^{16}$  is independently H, a  $C_1$ - $C_6$  alkyl, a benzyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, or a biologically compatible salt; and,

$R^{17}$  and  $R^{18}$ , where present, are independently H, a  $C_1$ - $C_6$  alkyl,  $C_1$ - $C_6$  carboxyalkyl, or a biologically compatible salt.

79. (New) A composition comprising a metal ion and a compound having the formula:



wherein  $R^1$  is  $-L-R_x$ ,  $-L-S_C$ ,  $-L-DYE$ ;  $C_1$ - $C_{18}$  alkyl or  $C_7$ - $C_{18}$  arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or

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heteroaryl ring system; or by  $-(\text{SO}_2)-\text{R}^{15}$ ,  $-(\text{SO}_2)-\text{O}-\text{R}^{15}$ ,  $-(\text{C}=\text{O})-\text{R}^{15}$ ,  $-(\text{C}=\text{O})-\text{O}-\text{R}^{16}$ ,  $-(\text{C}=\text{O})-\text{NR}^{17}\text{R}^{18}$ ; or by  $\text{C}_1-\text{C}_6$  alkylamino,  $\text{C}_2-\text{C}_{12}$  dialkylamino; or by  $\text{C}_1-\text{C}_6$  alkyl or  $\text{C}_1-\text{C}_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(\text{SO}_2)-\text{R}^{15}$ ,  $-(\text{SO}_2)-\text{O}-\text{R}^{15}$ ,  $-(\text{C}=\text{O})-\text{R}^{15}$ ,  $-(\text{C}=\text{O})-\text{O}-\text{R}^{16}$ ,  $-(\text{C}=\text{O})-\text{NR}^{17}\text{R}^{18}$ ;

$\text{R}^2$  is  $-\text{L}-\text{R}_x$ ,  $-\text{L}-\text{S}_c$ ,  $-\text{L}-\text{DYE}$ ;  $\text{C}_1-\text{C}_{18}$  alkyl or  $\text{C}_7-\text{C}_{18}$  arylalkyl, each of which is optionally substituted by halogen, azido, nitro, nitroso, amino, hydroxy, cyano, or by an aryl or heteroaryl ring system; or by  $-(\text{SO}_2)-\text{R}^{15}$ ,  $-(\text{SO}_2)-\text{O}-\text{R}^{15}$ ,  $-(\text{C}=\text{O})-\text{R}^{15}$ ,  $-(\text{C}=\text{O})-\text{O}-\text{R}^{16}$ ,  $-(\text{C}=\text{O})-\text{NR}^{17}\text{R}^{18}$ ; or by  $\text{C}_1-\text{C}_6$  alkylamino,  $\text{C}_2-\text{C}_{12}$  dialkylamino; or by  $\text{C}_1-\text{C}_6$  alkyl or  $\text{C}_1-\text{C}_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(\text{SO}_2)-\text{R}^{15}$ ,  $-(\text{SO}_2)-\text{O}-\text{R}^{15}$ ,  $-(\text{C}=\text{O})-\text{R}^{15}$ ,  $-(\text{C}=\text{O})-\text{O}-\text{R}^{16}$ ,  $-(\text{C}=\text{O})-\text{NR}^{17}\text{R}^{18}$ ;

wherein  $\text{R}^{15}$  is H,  $\text{C}_1-\text{C}_6$  alkyl,  $-\text{L}-\text{R}_x$ ,  $-\text{L}-\text{S}_c$ , or  $-\text{L}-\text{DYE}$ ;

$\text{R}^{16}$  is H, a  $\text{C}_1-\text{C}_6$  alkyl, a benzyl, alpha-acyloxyalkyl, t-butyldimethylsilyl, a biologically compatible salt,  $-\text{L}-\text{R}_x$ ,  $-\text{L}-\text{S}_c$ , or  $-\text{L}-\text{DYE}$ ;

$\text{R}^{17}$  is H,  $\text{C}_1-\text{C}_6$  alkyl,  $\text{C}_1-\text{C}_6$  carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt,  $-\text{L}-\text{R}_x$ ,  $-\text{L}-\text{S}_c$ , or  $-\text{L}-\text{DYE}$ ;

$\text{R}^{18}$  is H,  $\text{C}_1-\text{C}_6$  alkyl,  $\text{C}_1-\text{C}_6$  carboxyalkyl, an alpha-acyloxyalkyl, a t-butyldimethylsilyl, a biologically compatible salt,  $-\text{L}-\text{R}_x$ ,  $-\text{L}-\text{S}_c$ , or  $-\text{L}-\text{DYE}$ ;

or  $\text{R}^{17}$  and  $\text{R}^{18}$  taken in combination form a 5- or 6-membered aliphatic ring that optionally incorporates an oxygen atom;

L is a covalent linkage;

$\text{R}_x$  is an acrylamide, an activated ester of a carboxylic acid, an acyl azide, an acyl nitrile, an aldehyde, an alkyl halide, an anhydride, an aniline, an aryl halide, an azide, an aziridine, a boronate, a diazoalkane, a haloacetamide, a halotriazine, a hydrazine, an imido ester, an isocyanate, an isothiocyanate, a maleimide, a phosphoramidite, a reactive platinum complex, a silyl halide, a sulfonyl halide, or a thiol;

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Sc is an amino acid, a peptide, a protein, a polysaccharide, a nucleoside, a nucleotide, an oligonucleotide, a nucleic acid, a hapten, a psoralen, a drug, a hormone, a lipid, a lipid assembly, a synthetic polymer, a polymeric microparticle, a biological cell, a biotin, a silica or a virus;

DYE is a chemical moiety with an absorption maximum beyond 320 nm;

R<sup>7</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>8</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>9</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>10</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>11</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>12</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

R<sup>13</sup> is H, halogen, azido, nitro, nitroso, amino, cyano, -L-R<sub>x</sub>, -L-S<sub>C</sub>, -L-DYE, C<sub>1</sub>-C<sub>6</sub> alkyl or C<sub>1</sub>-C<sub>6</sub> alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy, -(SO<sub>2</sub>)-R<sup>15</sup>, -(SO<sub>2</sub>)-O-R<sup>15</sup>, -(C=O)-R<sup>15</sup>, -(C=O)-O-R<sup>16</sup>, or -(C=O)-NR<sup>17</sup>R<sup>18</sup>;

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$R^{14}$  is H, halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_x$ ,  $-L-S_c$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is itself optionally substituted by halogen, amino, hydroxy,  $-(SO_2)-R^{15}$ ,  $-(SO_2)-O-R^{15}$ ,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

or any two adjacent substituents  $R^7-R^{14}$ , taken in combination, form a fused six-membered benzo moiety, which is optionally substituted by halogen, azido, nitro, nitroso, amino, cyano,  $-L-R_x$ ,  $-L-S_c$ ,  $-L-DYE$ ,  $C_1-C_6$  alkyl or  $C_1-C_6$  alkoxy, each of which is optionally substituted by halogen, amino, hydroxy,  $-(C=O)-R^{15}$ ,  $-(C=O)-O-R^{16}$ , or  $-(C=O)-NR^{17}R^{18}$ ;

or any two adjacent substituents  $R^7-R^{14}$ , taken in combination with each other, and with the aromatic ring they are bound to, form a fused DYE;

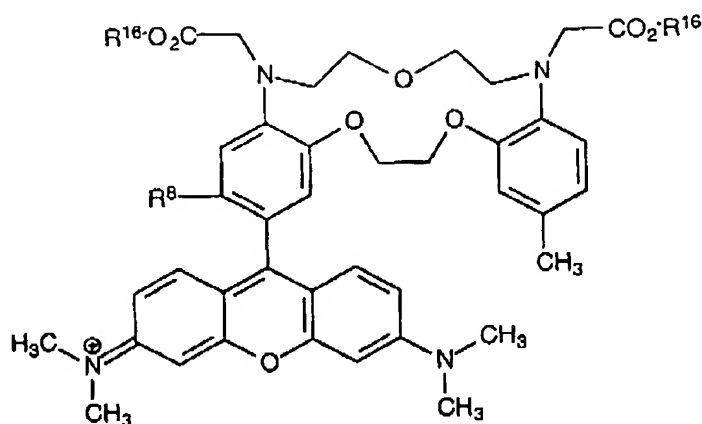
provided that the compound is substituted by at least one  $-L-DYE$ ,  $-L-R_x$ , or  $-L-S_c$  at  $R^1$ ,  $R^2$ ,  $R^4$ ,  $R^7$ ,  $R^8$ ,  $R^9$ ,  $R^{10}$ ,  $R^{11}$ ,  $R^{12}$ ,  $R^{13}$ , or  $R^{14}$ ; or at least two of  $R^7-R^{14}$ , taken in combination, form a fused DYE.

80. (New) The composition according to Claim 79, wherein the metal ion is  $Na^+$ ,  $K^+$ ,  $Ca^{2+}$ , or  $Zn^{2+}$ .
81. (New) The composition according to Claim 79, wherein DYE is an indole, a coumarin, a stilbene, a xanthene, an oxazine, a polyazaindacene, a benzofuran, a pyrene, an anthracene, a naphthalene, an acridine, a benzindole, an oxazole, a benzoxazole, a thiazole, a benzothiazole, a 4-amino-7-nitrobenz-2-oxa-1,3-diazole (NBD), a cyanine, a carbocyanine, a carbostyryl, a porphyrin, a salicylate, an anthranilate, an azulene, a perylene, a pyridine, a quinoline, a benzoxazine, a carbazine a phenalenone or a benzphenalenone.
82. (New) A method of detecting a target cationic metal ion in a sample, comprising:

- a) adding to the sample, in an amount sufficient to generate a detectable optical response when the target ion is present, a compound having the formula:

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and its salts, wherein

$R^8$ , is H or a  $C_1$ - $C_8$  alkoxy, which is optionally substituted by -  
( $C=O$ )- $O$ - $R^{16}$  or -( $C=O$ )- $NR^{17}R^{18}$ ;

$R^{16}$  is independently H, a  $C_1$ - $C_6$  alkyl, a benzyl, an alpha-  
acyloxyalkyl, a t-butyldimethylsilyl, or a biologically compatible  
salt; and,

$R^{17}$  and  $R^{18}$ , where present, are independently H, a  $C_1$ - $C_8$  alkyl,  
 $C_1$ - $C_6$  carboxyalkyl, or a biologically compatible salt;

- b) illuminating the sample to generate the detectable optical response whereby said  
target ion is detected.



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### CONCLUSION

In light of the above amendments, reconsideration and withdrawal of the outstanding objections and rejections are respectfully requested. All amendments are made in a good faith effort to advance the prosecution on the merits. Applicant respectfully submits that no amendments have been made to the pending claims for the purpose of overcoming any prior art rejections that would restrict the literal scope of the claims or equivalents thereof. Applicant reserves the right to subsequently take up prosecution of the claims originally filed in this application in continuation, continuation-in-part, and/or divisional applications.

It is submitted that this application is now ready for allowance. Early notice to this effect is solicited. If, in the opinion of the Examiner, a telephone conference would expedite the prosecution of the subject application, the Examiner is invited to call the undersigned at (541) 335-0203.

Respectfully submitted,

Date: March 3, 2005

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